Amendments to the Specification

Amend paragraph **[0017]** in the publication to read as follows:

[0017] It is not intended that any of the methods of the present invention be conducted in any particular order, as far as preparation of ~~pepsets~~ PEPSETS™ and differentiation of dendritic cells. For example, in some embodiments the ~~pepsets~~ PEPSETS™ are prepared before the dendritic cells are differentiated, while in other embodiments, the dendritic cells are differentiated before the ~~pepsets~~ PEPSETS™ are prepared, and in still other embodiments, the dendritic cells are differentiated and the ~~pepsets~~ PEPSETS™ are prepared concurrently. Thus, it is not intended that the present invention be limited to methods having these steps in any particular order.

Amend paragraphs **[0117]** and **[0119]** in the publication to read as follows:

[0117] In preferred embodiments, the methods provided by the present invention involve the use of dendritic cells as antigen-presenting cells, 15-mer peptides offset by 3 amino acids that encompass an entire protein sequence of interest, and $CD4^+$ T-cells obtained from the dendritic cell donors. T-cells are allowed to proliferate in a sample in the presence of the peptides (each peptide is tested individually) and differentiated dendritic cells. It is not intended that any of the methods of the present invention be conducted in any particular order, as far as preparation of the ~~pepsets~~ PEPSETS™ and differentiation of dendritic cells. For example, in some embodiments, the ~~pepsets~~ PEPSETS™ are prepared before the dendritic cells are differentiated, while in other embodiments, the dendritic cells are differentiated before the ~~pepsets~~ PEPSETS™ are prepared, and in still other embodiments, the dendritic cells are differentiated and the ~~pepsets~~ PEPSETS™ are prepared concurrently. Thus, it is not intended that the present invention be limited to methods having these steps in any particular order.

[0119]The present invention also provides methods for determining the immune response of a test population against a test protein, comprising the steps of: (a) preparing a ~~pepsets~~ PEPSETS™ from a test protein; (b) obtaining a plurality of solutions comprising human dendritic cells and a plurality of solutions of naïve human CD4+ and/or CD8+ T-cells, wherein the solutions of human dendritic cells and solutions of naïve human CD4+ and/or CD8+ T-cells

are obtained from a plurality of individuals within the test population; (c) differentiating the dendritic cells to produce a plurality of solutions comprising differentiated dendritic cells; (d) combining the plurality of the solutions of differentiated dendritic cells and the solutions of naïve CD4+ and/or CD8+ T-cells with the ~~pepsets~~ PEPSETS™, wherein each of the solutions of differentiated dendritic cells and the solutions of naïve CD4+ and/or CD8+ T-cells are from one individual within the test population are combined; (e) measuring proliferation of the T-cells in step (d), to determine the responses to each peptide in the ~~pepsets~~ PEPSETS™; (g) compiling the responses of the T-cells in step (e) for the test protein; (h) determining the structure value of the compiled responses of step (g) for the test protein; and (i) determining the level of exposure of the plurality of individuals to the test protein. In some preferred embodiments, the ~~pepsets~~ PEPSETS™ comprise peptides of about 15 amino acids in length, while in some particularly preferred embodiments each peptide overlaps adjacent peptides by about 3 amino acids. However, it is not intended that the peptides within the ~~pepsets~~ PEPSETS™ be limited to any particular length nor overlap, as other peptide lengths and overlap amounts find use in various embodiments of the present invention. In some embodiments, at least two test proteins are tested. In some preferred embodiments, the level of exposure of the plurality of individuals to the test protein is compared. In some particularly preferred embodiments, the test protein is modified to produce a variant protein that exhibits a reduced immunogenic response in the test population. The present invention also provides means to categorize proteins based on both their background percent response and their structure values. Thus, in some further embodiments, the proteins analyzed are categorized and/or ranked according to their background percent response and structure values.

Amend paragraph **[0124]** in the publication to read as follows:

[0124] In some embodiments, the present invention provides methods for ranking the relative immunogenicity of a first protein and at least one additional protein, comprising the steps of: (a) preparing a first ~~pepsets~~ PEPSETS™ from a first protein and preparing at least one additional ~~pepset~~ PEPSET™ from each of the additional proteins, wherein each of the ~~pepsets~~ PEPSETS™ (b) obtaining from a single human blood source a solution comprising dendritic cells and a solution of naïve CD4+ and/or CD8+ T-cells; (c) differentiating the dendritic cells to produce a solution of differentiated dendritic cells; (d) combining the solution of differentiated dendritic

cells and the naïve CD4+ and/or CD8+ T-cells with the first ~~pepsets~~ PEPSETS™; (e) combining the solution of differentiated dendritic cells and the naïve CD4+ and/or CD8+ T-cells with each of the ~~pepsets~~ PEPSETS™ from the additional proteins; measuring proliferation of the T-cells in steps (d) and (e), to determine the responses to each peptide in the first and additional ~~pepsets~~ PEPSETS™; (g) compiling the responses of the T-cells in step (f) for the first protein and the additional proteins; (h) determining the structure value of the compiled responses of step (g) for the first protein and the additional proteins; and (i) comparing the structure value obtained for the first protein with the structure value for the additional proteins to determine the immunogenicity ranking of the first protein and the additional proteins. In some preferred embodiments, the ~~pepsets~~ PEPSETS™ comprise peptides of about 15 amino acids in length, while in some particularly preferred embodiments each peptide overlaps adjacent peptides by about 3 amino acids. However, it is not intended that the peptides within the ~~pepsets~~ PEPSETS™ be limited to any particular length nor overlap, as other peptide lengths and overlap amounts find use in various embodiments of the present invention.

Amend paragraphs **[0126]** and **[0127]** in the publication to read as follows:

[0126] The present invention also provides methods for ranking the relative immunogenicity of two proteins, wherein the second protein is a protein variant of the first protein, comprising the steps of: (a) preparing a first ~~pepsets~~ PEPSETS™ from a first protein and a second ~~pepsets~~ PEPSETS™ from a second protein; (b) obtaining from a single human blood source a solution comprising dendritic cells and a solution of naïve CD4+ and/or CD8+ T-cells; (c) differentiating the dendritic cells to produce a solution of differentiated dendritic cells; (d) combining the solution of differentiated dendritic cells and the naïve CD4+ and/or CD8+ T-cells with the first ~~pepsets~~ PEPSETS™; (e) combining the solution of differentiated dendritic cells and the naïve CD4+ and/or CD8+ T-cells with the second ~~pepsets~~ PEPSETS™; (f) measuring proliferation of the T-cells in steps (d) and (e), to determine the responses to each peptide in the first and second ~~pepsets~~ PEPSETS™; (g) compiling the responses of the T-cells in step (f) for the first protein and the second protein; (h) determining the structure value of the compiled responses of step (g) for the first protein and the second protein; (i) comparing the structure value obtained for the first protein with the structure value for the second protein to determine the immunogenicity ranking of the first protein and the second protein. In some embodiments, the second protein is ranked as

less immunogenic than the first protein, while in alternative embodiments, the first protein is ranked as less immunogenic than the second protein. In some preferred embodiments, the ~~pepsets~~ PEPSETS™ comprise peptides of about 15 amino acids in length, while in some particularly preferred embodiments each peptide overlaps adjacent peptides by about 3 amino acids. However, it is not intended that the peptides within the ~~pepsets~~ PEPSETS™ be limited to any particular length nor overlap, as other peptide lengths and overlap amounts find use in various embodiments of the present invention. In still further embodiments, a positive response against the first protein comprises a stimulation index value between about 2.7 and about 3.2, while in other embodiments, a positive response against the second protein comprises a stimulation index value between about 2.7 and about 3.2. In additional embodiments, a positive response against the first protein comprises a stimulation index value between about 2.7 and about 3.2 and a positive response against the second protein comprises a stimulation index value between about 2.7 and about 3.2. In still further embodiments, the proliferation of the T-cells in steps (d) results in a stimulation index of about 2.95 or greater and the proliferation of the T-cells in steps (e) results in a stimulation index of about 2.95 or greater. In some particularly preferred embodiments, at least one additional human blood source is used in step (b). In some additional particularly preferred embodiments, the structure values obtained for each of the human blood sources and the proteins are compared. In some embodiments, the second protein comprises a reduction of at least one prominent region in the first protein. In further embodiments, the proliferation of the T-cells in step (e) is at a background level. In some particularly preferred embodiments, the structure values obtained for each of the human blood sources and the proteins are compared. The present invention also provides means to categorize proteins based on both their background percent response and their structure values. Thus, in some further embodiments, the proteins analyzed are categorized and/or ranked according to their background percent response and structure values.

[0127] The present invention also provides methods for ranking the relative immunogenicity of a first protein and at least one variant protein, comprising the steps of: (a) preparing a first ~~pepset~~ PEPSET™ from a first protein and ~~pepsets~~ PEPSETS™ from each of the variant proteins; (b) obtaining from a single human blood source a solution comprising dendritic cells and a solution of naïve CD4+ and/or CD8+ T-cells; (c) differentiating the dendritic cells to produce a solution

of differentiated dendritic cells; (d) combining the solution of differentiated dendritic cells and the naïve CD4+ and/or CD8+ T-cells with the first pepset; (e) combining the solution of differentiated dendritic cells and the naïve CD4+ and/or CD8+ T-cells with each pepset prepared from each of the variant proteins; (f) measuring proliferation of the T-cells in steps (d) and (e), to determine the responses to each peptide in the first and second ~~pepsets~~ PEPSETS™; (g) compiling the responses of the T-cells in step (f) for the first protein and the variant protein(s); (h) determining the structure value of the compiled responses of step (g) for the first protein and the variant protein(s); and (i) comparing the structure value obtained for the first protein with the structure value for the variant protein(s) to determine the immunogenicity ranking of the first protein and the variant proteins. In some preferred embodiments, the ~~pepsets~~ PEPSETS™ comprise peptides of about 15 amino acids in length, while in some particularly preferred embodiments each peptide overlaps adjacent peptides by about 3 amino acids. However, it is not intended that the peptides within the ~~pepsets~~ PEPSETS™ be limited to any particular length nor overlap, as other peptide lengths and overlap amounts find use in the present invention. In some preferred embodiments, at least one of the variant proteins is ranked as less immunogenic than the first protein, while in other embodiments, the first protein is ranked as less immunogenic than at least one of the variant proteins. In further embodiments, a positive response against the first protein comprises a stimulation index value between about 2.7 and about 3.2, while in other embodiments, a positive response against a variant protein comprises a stimulation index value between about 2.7 and about 3.2. In additional embodiments, a positive response against the first protein comprises a stimulation index value between about 2.7 and about 3.2 and a positive response against a variant protein comprises a stimulation index value between about 2.7 and about 3.2. In still further embodiments, the proliferation of the T-cells in steps (d) results in a stimulation index of about 2.95 or greater and the proliferation of the T-cells in steps (e) results in a stimulation index of about 2.95 or greater. In some particularly preferred embodiments, at least one additional human blood source is used in step (b). In some additional particularly preferred embodiments, the structure values obtained for each of the human blood sources and the proteins are compared. In some embodiments, the variant protein comprises a reduction of at least one prominent region in the first protein. In further embodiments, the proliferation of the T-cells in step (e) is at a background level. In some preferred embodiments, the proliferation of the T-cells in step (e) for at least one variant protein is at a background level. In some

particularly preferred embodiments, the structure values obtained for each of the human blood sources and the proteins are compared. In further embodiments, at least one additional human blood source is used in step (b). The present invention also provides means to categorize proteins based on both their background percent response and their structure values. Thus, in some further embodiments, the proteins analyzed are categorized and/or ranked according to their background percent response and structure values.

Amend paragraphs **[0132]** and **[0134]** in the publication to read as follows:

[0132] Peripheral mononuclear blood cells (PBMCs) (stored at room temperature, no older than 24 hours) were prepared for use as follows. PBMCs were isolated from buffy coat material by centrifuging over an underlay of ~~Lymphoprep~~LYMPHOPREP™ at 1000 xg for 30 minutes. The interface layer was collected and washed and counted using the ~~Cell-Dyn~~ CELL-DYN® 3700 System (Abbott). Then, suspensions containing 10^8 PBMCs resuspended in 30 ml of AIM-V (Invitrogen) were prepared and the cells were allowed to adhere to plastic T-75 culture flasks for two hours. The remainder of the cells were frozen at 5 x 10^7 cells/ml in 90% FCS (Gibco/BRL) and 10% DMSO (Sigma).

[0134] Autologous CD4+ T-cells were prepared from frozen aliquots of PBMCs. After thawing and washing in DPBS, CD4+ T-cells were isolated using a commercially available CD4 negative selection kit (Dynal), according to the manufacturer's instructions. Cells were counted using the Abbott ~~Cell-Dyn~~ CELL-DYN® 3700 System. The purity obtained using these methods was generally found to be greater than 90%.

Amend paragraph **[0136]** in the publication to read as follows:

[0136] After 5 days of incubation at 37° C, 5% CO_2, the cultures were pulsed with 0.25 μCi/well tritiated thymidine (Perkin Elmer). After a subsequent 24 hours of incubation, plates were harvested and assessed for incorporation of the tritiated thymidine (*i.e.*, T-cell proliferation) using a Wallac ~~Microbeta~~ MICROBETA® TriLux liquid scintillation counter (Perkin Elmer).